8 to page 80, line 10, for example. Support for Claim 59 can be found on page 34, lines 14-34, for example. See also page 33, lines 4-20 and page 33, line 32 to page 34, line 3 as support for all the added claims.

Rejection of Claims 15-17 and 49-52 Under 35 U.S.C. § 102(b) (Item 5, Page 3 of Office Action)

Claims 15-17 have been rejected under 35 U.S.C. § 102(b) "as being anticipated by Stamler *et al.*, WO 93/09806 (5/93)." "The reference teaching of the use of S-nitrosylhemoglobin..." is said to "...envisage (e.g., anticipate) the use of either the oxygenated or deoxygenated forms of hemoglobin e.g., SNO-Hb[FeII]O_2 and/or SNO-Hb[FeIII] as presently claimed.

Stamler *et al.* (WO 93/09806) disclose *S*-nitroso-proteins, in particular, *S*-nitroso-tPA, *S*-nitroso-BSA, *S*-nitroso-cathepsin B, *S*-nitroso-lipoprotein and *S*-nitroso-immunoglobulin, and methods for producing the same, using NO or $NaNO_2$ as the reagent, under acidic conditions. They also report a method which they claim results in the synthesis of *S*-nitroso-hemoglobin.

As explained in the Declaration of Jonathan S. Stamler, M.D. Under 37 C.F.R. § 1.132, no form of SNO-Hb is enabled. No other form of nitrated or nitrosated hemoglobin is mentioned in WO 93/09806 as being useful in a method of therapy for any kind of medical disorder.

Rejection of Claims 15-17 and 49-52 Under 35 U.S.C. § 103(a) (Item 7, Page 4 of Office Action)

Claims 15-17 and 49-52 have been rejected under 35 U.S.C. § 103(a) "as obvious over Stamler *et al.*, WO 93/09806 (5/93) and Kaesenmeyer, U.S. Pat. No. 5,543,430 (8/96; filed 10/94)."

The teachings of Stamler et al. (WO 93/09806) have been described above.

Kaesenmeyer (U.S. Patent No. 5,543,430) describes a method for treating cardiovascular diseases using L-arginine and a vasodilator, a compound "which when administered to a subject is converted biologically to nitric oxide (NO) which is a pharmacologically active metabolite." See column 1, lines 21-24. *S*-nitrosothiols are mentioned as being vasodilators. See column 1, line 49. Nitrates are also mentioned as a source for the production of NO. See column 6, lines 19-23, and lines 44-47. Kaesenmeyer does not mention any form of hemoglobin, or any nitrosoprotein. It was thought at the time of the filing of the Kaesenmeyer application (October

5, 1994), that hemoglobin would *block* nitric oxide activity by acting as a scavenger of nitric oxide.

The Examiner states that "Stamler *et al.* generally teach the use of NO donor nitrosylated protein compounds (e.g., especially *S*-nitrosylated) for use in relaxing smooth muscle and inhibiting platelet aggregation." Contrary to what might have been predicted from the *S*-nitrosoproteins successfully produced as described in WO 93/09806, SNO-hemoglobin, as studied in the standard bioiassay, in the presence of oxygen, does not act as a vasodilator (see, for example, Figure 4A and page 68, lines 3-14 of the specification).

The Stamler *et al.* published patent application does not and cannot teach the use of NO donor nitrosylated *hemoglobin* for use in relaxing smooth muscle, for inhibiting platelet aggregation, or for anything else. As can be concluded from the Declaration of Jonathan S. Stamler, M.D. Under 37 C.F.R. § 1.132, being filed concurrently with this Amendment, no form of nitrosated, nitrated, or nitrosylated hemoglobin is enabled by the disclosure of WO 93/09806. The description of a method to produce *S*-nitrosohemoglobin appearing in WO 93/09806 was followed in an attempt to reproduce the method, as explained in the Declaration, and did not produce *S*-nitrosohemoglobin.

Therefore, from the combination of cited references, one of ordinary skill in the art might seek an *S*-nitrosothiol of some kind to be used as a vasodilator, and might turn to an *S*-nitrosoprotein as a candidate vasodilator. However, from other art known at the time of filing the priority application (see, for example, Greenburg, A.G., and H.W. Kim, *Art. Cells, Blood Subs. and Immob. Biotech., 23(3)*:271-276, 1995; cited as reference AZ3), one would know that hemoglobin was thought to act as a scavenger of nitric oxide. This would discourage investigations into the use of any form of hemoglobin as a vasodilator or as an inhibitor of platelet activation. Presented with a description of a method to produce SNO-hemoglobin in WO 93/09806, one of skill in the art woud be unable to follow it to produce SNO-hemoglobin.

Rejection of Claims 15-17 and 49-52 Under 35 U.S.C. § 103(a) (Item 8, Page 5 of Office Action)

Claims 15-17 and 49-52 have been rejected under 35 U.S.C. § 103(a) "as being unpatentable over Stamler *et al.*, WO 93/09806 (5/93) and Hsia, U.S. Pat. No. 5,591,710 (1/97; filed 8/94)."

The Examiner states that "Stamler et al. disclose the use of a nitrosated hemoglobin (e.g., see definition of 'nitrosylation' (page 14, lines 7-11 [of WO 93/09806]) and specific disclosure of S-nitrosohemoglobin) for promoting vasodilation and platelet inhibition and to treat/prevent cardiovascular disorders (e.g., see page 19, lines 22-25)."

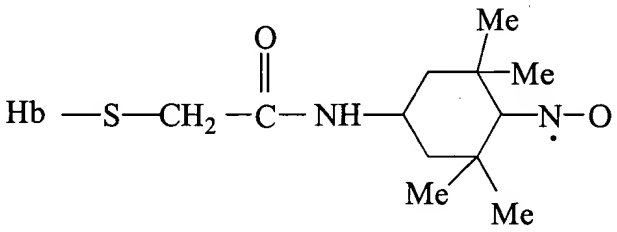
The Stamler *et al.* published patent application WO 93/09806 does not and cannot teach the use of a nitrosated hemoglobin for promoting vasodilation and platelet inhibition and for treating or preventing cardiovascular disorders. As explained in the Declaration of Jonathan S. Stamler, M.D. Under 37 C.F.R. § 1.132, the making and using of *S*-nitrosohemoglobin is not taught by the disclosure of WO 93/09806. The description of a method to produce *S*-nitrosohemoglobin in WO 93/09806 was recently followed, as explained in the Declaration, and did not produce *S*-nitrosohemoglobin.

At the time of the invention, another form of nitrosated hemoglobin was known, nitrosylhemoglobin (NO-Hb), having the NO bound at the heme iron. However, there is no suggestion in either of the cited references that this form of hemoglobin would be useful in the prevention or inhibition of platelet aggregation. It was known from published studies, at the time of the invention, that the affinity of the heme iron for NO is extremely high, and that NO-Hb is extremely stable. Therefore, NO-Hb was not considered to be a possible donor of NO useful in any method of therapy where one would want to provide NO to cells or tissues. See, for example, Greenburg, A.G., and H.W. Kim, *Art. Cells, Blood Subs. and Immob. Biotech., 23(3)*:271-276, 1995 (cited as reference AZ3). See especially the last paragraph on page 272 and the first paragraph of page 273, wherein the high affinity of NO for the heme iron is discussed, and it is said that "there appears to be no relationship between HbNO formation and the ability to induce vasodilation."

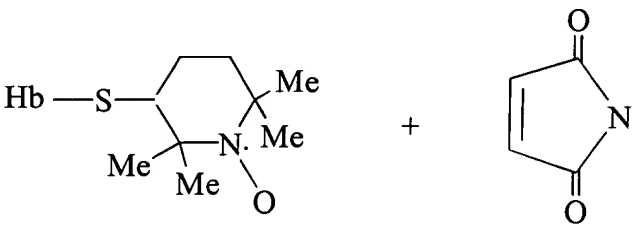
Hsia (US 5,591,710) discloses hemoglobin conjugated with nitroxides, where "nitroxide" is narrowly defined (see column 9, lines 54-60 *et seq.*) as stable nitroxide free radicals, their precursors and their derivatives, in which a nitrogen atom is attached to two carbon atoms. NItroxides are not nitric oxide and are not nitric oxide donors. Hsia also discloses methods of making these modified hemoglobins, and intended uses for the modified hemoglobins as therapeutic agents to detoxify superoxide anion. "Nitroxide" as defined in Hsia includes compounds having N bound to an R group (see top of column 11) or N specifically bound to adjacent carbon atoms (column 9, lines 61-67). Hsia teaches that these nitroxides may be

covalently attached to hemoglobin at one or more sites on the hemoglobin molecule, for example, -SH, -NH_2 or -COOH groups.

More specifically, for example, in column 17, Hsia teaches the use of nitroxides with functional groups which are reactive with thiols to attach *the entire nitroxide*, including R group, to the thiols of hemoglobin, not nitric oxide. The product of the reaction between the TEMPO (column 17, lines 10-20) nitroxide and hemoglobin is:



The products of the reaction between the PROXYL nitroxide (column 17, lines 22-30) and hemoglobin are:



This process is not nitrosation, which is defined in the specification at page 37, line 17 to page 38, line 7. It is clear that by this definition, a nitrogen atom must be covalently attached to the nucleophilic atom in the reaction, and the product is not a radical. See page 37, lines 25-28 in the specification, giving examples of the products of the nitrosation reactions defined. What is attached to hemoglobin by Hsia is a nitroxide, at a nucleophilic center, to form a group of the

type ~S-C=N-O·. The reaction of Hsia is no more a nitrosation than it is a methylation. (The TEMPO and PROXYL reagents, for example, have methyl groups within their structures.) The reagents of Hsia cannot by any plausible chemical reaction donate NO. The nitroxides of Hsia are in no way "NO-donating compounds" and would not be expected by one of ordinary skill in the art to behave as NO donors in a method of therapy. Rather, the hemoglobins of Hsia are *nitroxidated*, and act as free radical scavengers.

The purpose of the modified hemoglobins described in Hsia is to give "superoxide oxidase" activity to a hemoglobin-based blood substitute that is to be stored for some period of time before use. See column 12, lines 13-33. The nitroxidated hemoglobins of Hsia do not have the ability to release and deliver NO biological equivalents to tissues, as do the nitrosated or nitrated hemoglobins of the claims. Therefore, Hsia does not teach or suggest the invention. Hsia does not and cannot provide instruction or motivation for one of skill in the art to produce nitrosated or nitrated hemoglobin, or a method of therapy in which platelet aggregation is inhibited.

Combining WO 93/09806 with Hsia, one of ordinary skill in the art might be motivated, from WO93/09806, to try to produce a nitrosated hemoglobin, other than NO-Hb, and to use it in a method of inhibiting platelet aggregation. Considering the teachings of the prior art that hemoglobin acts as a scavenger of nitric oxide, it is difficult to find such motivation. Even assuming such motivation is found, instruction in making and using a nitrosated hemoglobin is not provided by either of the cited references. See the Declaration of Jonathan S. Stamler, M.D. Under 37 C.F.R. § 1.132 regarding the description in WO 93/09806. The chemistry taught in Hsia is not applicable to producing NO donors.

Provisional Rejection of Claims 15-17 Under 35 U.S.C § 101 (Item 10, Page 6 of Office Action)

Claims 15-17 have been provisionally rejected under 35 U.S.C § 101 as claiming the same invention as that of claims 30-32 of copending Application No. 08/796,164.

Claims 30-32 of 08/796,164 have been canceled in an Amendment After Final Action faxed to the United States Patent and Trademark Office on August 30, 1999, thereby obviating the rejection.

Provisional Rejection of Claims 49-52 -- Obviousness-Type Double Patenting (Item 11, Page 7 of Office Action)

Claims 49-52 have been provisionally rejected as being unpatentable over Claims 10-15 and 30-32 of copending Application No. 08/796,164. The Examiner states:

> Although the conflicting claims are not identical, they are not patentably distinct from each other because the '164 application discloses the use of S-nitrosylated hemoglobins generally (e.g., claims 30-32) in the presently claimed invention and additionally discloses the making and formulation of a specific S-nitrosylated hemoglobin e.g., SNO-Hb[FeII]O_2. Accordingly, the selection of the specific S-nitrosylated hemoglobin e.g., SNO-Hb[FeII]O_2 and its reduced obvious variant e.g., SNO-Hb[FeIII] for use in the '164 claim 30-32 methods would have been obvious to one of ordinary skill at the time of applicant's invention.

Claims 30-32 of 08/796,164 have been canceled in an Amendment After Final Action faxed to the United States Patent and Trademark Office on August 30, 1999.

> In determining whether a nonstatutory basis exists for a double patenting rejection, the first question to be asked is -- does any claim in the application define an invention that is merely an obvious variation of an invention claimed in the patent? If the answer is yes, then an "obvious-type" nonstatutory double patenting rejection may be appropriate. M.P.E.P. sec. 804 (B) (1.)

Claims 49-52 of the subject application are drawn to methods for treating a disorder resulting from platelet activation or adherence and methods for preventing thrombus formation. Claims 10-15 of copending application 08/796,164 are drawn to compositions comprising SNO-Hb[FeII]O_2, compositions comprising SNO-Hb[FeII], methods for making SNO-Hb[FeII]O_2 and methods for making SNO-Hb[FeII].

Here, Claims 49-52 in no way define an invention that is an obvious variation of an invention claimed in Claims 10-15 of 08/796,164. Claims 10 and 13 are drawn to compositions. The methods of Claims 11, 12, 14 and 15 of 08/796,164 -- all methods of producing SNO-hemoglobin --do not have a similar object or similar steps to those of Claims 49-52 -- all drawn to methods of therapy.

CONCLUSION

The Examiner is respectfully requested to consider the above amendments and remarks, and to withdraw the rejections. If the Examiner feels that a telephone conference would expedite prosecution of this case, the Examiner is invited to call the undersigned attorney.

Respectfully submitted,

HAMILTON, BROOK, SMITH & REYNOLDS, P.C.

By Carol A. Egner

Carol A. Egner
Registration No. 38,866
Telephone (781) 861-6240
Facsimile (781) 861-9540

Lexington, Massachusetts 02421-4799
Dated: September 2, 1994